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Mature Larvae of Calliopsine Bees: *Spinoliella*, *Callonychium*, and *Arhysosage* Including Biological Notes, and a Larval Key to Calliopsine Genera (Hymenoptera: Apoidea: Andrenidae: Panurginae)

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ABSTRACT

The known mature larvae of andrenid genera *Spinoliella*, *Callonychium*, and *Arhysosage* are described and compared with one another and with those of other Calliopsini. On the basis of both larval and adult anatomy, these three genera represent a distinctive clade, the "*Spinoliella* clade." The larvae are characterized by conspicuously enlarged and oddly shaped, paired dorsal tubercles on the pro- and mesothorax, while the paired dorsal tubercles of most other body segments are erect, elongate, and slender. All dorsal tubercles bear fine setae. The larvae were collected on field trips to South America over a 35 year period. Associated field notes are reported, treating information on nesting ecology, nest structure and dimensions, provisions, larval behavior, associated cleptoparasites, and mating behavior. Also presented is a preliminary taxonomic key to the genera of Calliopsini based on mature larvae.

INTRODUCTION

Mature larvae of three genera, *Spinoliella*, *Callonychium*, and *Arhysosage* uniquely exhibit exaggerated paired dorsal pro- and mesothoracic tubercles compared with other tribal members. The exaggeration involves increase in size, modification of shape, and orientation of these tubercles compared with the paired dorsal tubercles on the remaining body segments and implies some

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special function since such extreme enlargement and backward tilting of the prothoracic tubercles does not occur elsewhere in the Andrenidae. The paired dorsal tubercles of other body segments are strongly erect and substantially more slender than the preceding tubercles, but, like the tubercles of the pro- and mesothorax, bear setose apices. The suggestion that this combination of paired dorsal body tubercles (enlarged, oddly oriented and shaped pro- and mesothoracic tubercles followed by slender, strongly erect body tubercles) may be an apomorphy is supported by Ruz's (1991) phylogenetic analysis of the Calliopsini and related tribes based on adult morphology in which the three genera are grouped as a clade sister to *Acamptopoeum* and *Calliopsis* s.l. referred to hereafter as the "*Spinoliella* clade." As the current study shows below, other larval features also demonstrate the close relationship of these three genera.

The key included here distinguishes known mature larvae of these three genera from those of other genera of the Calliopsini, i.e., *Acamptopoeum* and *Calliopsis* (nest and larva of *Litocalliopsis* unknown), as recognized by Michener (2007). A larval description of the clade is presented, as are illustrations of larvae of the three genera including two subgenera of *Callonychium*. The larvae of individual taxa are compared in the Diagnosis of each taxon.

Specimens used in this study have been collected over 35 years. In the process of excavating nests, information on habitat, nest structure, nest associates, provisions, and occasionally mating behavior have come to light. These data, including many novel observations, are presented under Biological Notes for each of the species and are summarized in Discussion.

Of the genera treated here, Claude-Joseph (1926) illustrated and briefly described mature larvae of *Spinoliella maculata* (Spinola) (as *Camptopaeum maculatum*) and *S. herbsti* (Friese) (as *Camptopaeum Herbsti* [sic] Friese). The larval form of *Arhysosage* has not been described before, nor has its nesting biology, except for its association with the cleptoparasite *Caenoprosopis crabronina* Holmberg (Apidae: Nomadinae) (Rozen and Roig-Alsina, 1991). Larvae of *Callonychium* have not been treated in the past, and nesting biologies have gone unreported, although Toro (1985) discussed the biomechanics of the mating behavior of *C.* (*Paranychium*) *chilense* Friese, and Cure and Wittmann (1990) commented on *C.* (*C.*) *petuniae* Cure and Wittmann flying in copulo while visiting flowers of *Petunia*. The following accounts are presented to further our understanding of the anatomy of larvae and the relationships of the genera. Further, it is hoped that they will stimulate inquiries into the adaptive function of the enlarged and modified paired dorsal pro- and mesothoracic tubercles and the slender, erect posterior dorsal tubercles, as outlined in the Discussion.

Larval exemplars of the other currently recognized genera of Calliopsini have been described, as follows: *Acamptopoeum* (Claude-Joseph, 1926; Rozen and Yanega, 1999) and *Calliopsis* (Michener, 1953; Rozen, 1958, 1963, 1966, 2008). The following key is based on specimens described in these papers as well as herein.

METHODS AND TERMINOLOGY

Larvae used in this study had been fixed and maintained in Kahle's solution from when found until selected for study. They were then examined and illustrated, after which heads and

TABLE 1. Comparative Nest Statistics of *Spinoliella*, *Callonychium*, and *Arhysosage*. Symbol ++ = exact number unknown but more than one. For explanation of double rows, see text.

Species	Burrow diameter (mm)	N	Cell depth (cm)	N	Cell length (mm)	N	Cell diameter (mm)	N	Egg length (mm)	N	Egg diameter (mm)	N	Food sphere (mm)	N
Spinoliella herbsti	4.0	1	7.5–12.0		12.0–13.0	4	5.5–6.0	4					3.75–4.25	3
Spinoliella maculata	5.0	1	8.0–11.0	++	15.0	1	6.8	1	2.05	1	0.50	1	3.4–4.2	3
Callonychium (C.) flaviventre	3.0–4.0	7	5.0–11.0	6	7.0–9.0[a] 9.2–9.3[b]	5 3	5.0–5.5[a] 5.3–5.5[b]	5 4	1.70–1.75	2	0.375	2	3.2–3.5[a] 3.3–3.95[b]	4 9
Callonychium (C.) petuniae	3.0	++	4.0–7.5	8	6.0–7.0	4	4.0–4.5	5	1.45–1.60	2	0.40	2	2.1–2.6	4
C. (Paranychium) minutum	1.5–2.0	9	6.0–9.0	11	5.0–7.0	13	2.7–3.2	13	0.93–1.13	5	0.25–0.28[c]	4	1.63–1.80	13
C. (Paranychium) n. sp.	2.0 2.0	4 3	14–20 4.0 (3–6)	6 4	4.5 5.0–5.5	1 2	3.0 3.0–3.3	1 2	1.0	1	0.28	1	1.73–1.75 1.8	2 2
Arhysosage flava	4.0–4.5	6	9.0–11.0	4	10.0	2	5.7–6.0	3					4.2–4.3	3
Arhysosage bifasciata	4.0–5.0	6	8.0–14.0	7	14.0–15.0	4	6.0–7.2	6	2.45	1	0.5	1	4.2–4.4	3

[a] Amaichá del Valle site.
[b] 8 km SW Ticucho site.
[c] Another recorded datum of 0.13 mm is assumed to be a lapsus.

bodies were separated and cleared of tissue by boiling in an aqueous solution of sodium hydroxide. After being washed in water, they were transferred to 70%–75% ethanol, stained with Chlorazol Black E, washed in ethanol, and submerged in glycerin on well slides for microscopic scrutiny and eventual storage.

In the following key and descriptions references are made to "midbody paired dorsal tubercles." These are the paired dorsal tubercles of the metathorax and at least abdominal segments 1–4. Those of a good many of the following abdominal segments are similar though decreasing in size toward the abdominal apex.

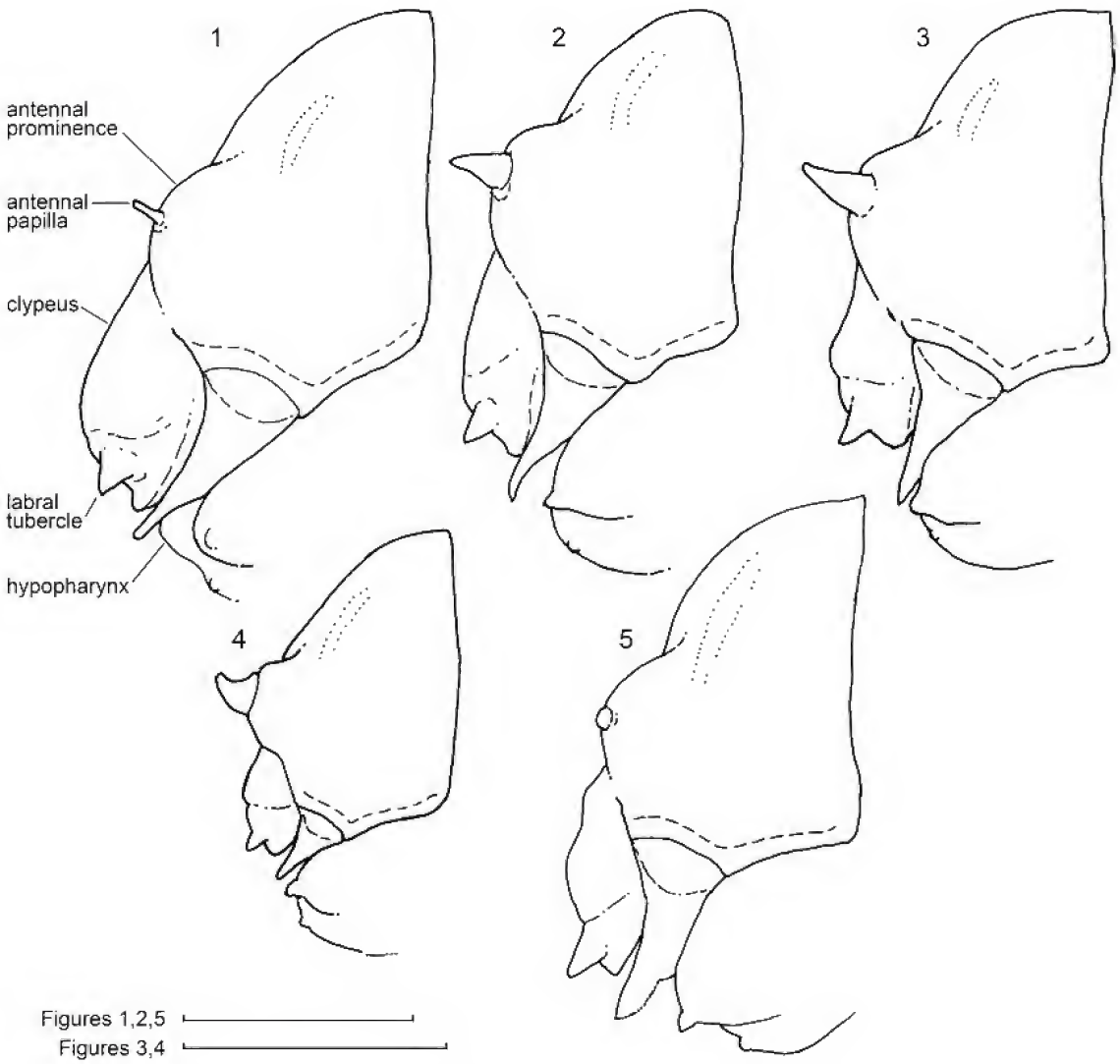
In nest descriptions "no visible special cell lining" is often mentioned. This means that the inner surface of a cell was approximately the same hue and tone as the surrounding soil, was not more (or less) reflective than the soil particles elsewhere, and/or did not exhibit a surface film, thereby suggesting that the female bee had provided no further treatment of the surface beyond smoothing it.

Nest statistics (burrow diameter, cell depth, length, and diameter) as well as egg dimensions and food-sphere diameters are presented in table 1 for each species.

Key to Genera of Calliopsini Based on Their Mature Larvae

1. Midbody paired dorsal tubercles low, with apices not attenuated; paired dorsal tubercles of prothorax not longer and at most only slightly larger than midbody paired tubercles (Rozen, 1958: figs. 53–62; Rozen and Yanega, 1999: figs. 4, 5; Rozen, 2008; figs. 8, 9); antennal papilla small, height less than basal diameter (Rozen 1958: figs. 4–27; Rozen and Yanega, 1999: figs. 6, 7; Rozen, 2008: figs. 10, 14, 18, 19). . . *Acamptopoeum* and *Calliopsis*
– Midbody paired dorsal tubercles slender, attenuated (figs. 6, 8, 10, 12, 14, 16, 17); paired dorsal tubercles of prothorax substantially larger than midbody paired dorsal tubercles (figs. 6–18); antennal papilla tending to be large and as long as, or longer than, basal diameter (figs. 1–4) except in *Arhysosage flava* (fig. 5). *Spinoliella* clade 2
2(1). Antennal papilla small, inconspicuous, slightly shorter than basal diameter, its apex broadly rounded (fig. 5); apices of mesothoracic paired dorsal tubercles directed laterad with extreme apices turning backward (figs. 17, 18); ventral apical mandibular edge finely serrate (figs. 30, 31) . . . . . *Arhysosage flava* Moure
– Antennal papilla larger (sometimes very large), tending to be more conspicuous, as long as, or longer than, basal diameter (figs. 1–4), its apex gradually, evenly narrowing in lateral view (figs. 1–4); apices of mesothoracic paired dorsal tubercles straight, pointing away from body center (figs. 7, 9, 11, 13, 15); ventral apical mandibular edge smooth (fig. 29). 3
3(2). Mesothoracic paired tubercles distinctly larger than metathoracic tubercles, projecting posteriorly away from prothoracic tubercles (figs. 6, 8); antennal papilla slender, far less massive than labral tubercle (fig. 1) . . . . . .*Spinoliella herbsti* (Friese)
S. maculata (Spinola)
– Mesothoracic paired tubercles more similar in size to metathoracic paired tubercles, either projecting away from thorax in lateral view (figs. 14, 16) or projecting forward toward prothoracic tubercles, so that they nearly touch in lateral view (figs. 10, 12); antennal papilla as massive as, or more massive than, labral tubercle (figs. 2–4). *Callonychium* s.l. 4
4(3). Mesothoracic paired tubercle projecting vertically away from body in lateral view (figs. 14, 16); antennal papilla curving upward in lateral profile (fig. 4) . . . . . .*Callonychium* (*Paranychium*) *minutum* (Friese)
Callonychium (*Paranychium*) n. sp.
– Mesothoracic paired tubercles projecting forward toward prothoracic tubercles so that they nearly touch one another in lateral view (figs. 10, 12); antennal papilla nearly (*C. petuniae*, fig. 3) or actually (*C. flaviventre,* fig. 2) straight in lateral profile . . . . . *Callonychium* (*C.*) *flaviventre* (Friese)
C. (*C.*) *petuniae* Cure and Wittmann

This key is based on the mature larvae of the following in addition to the species identified in the key above: *Acamptopoeum prinii* (Holmberg), *Calliopsis* (*Calliopsis*) *andreniformis* Smith, *C.* (*Calliopsima*) *rozeni* Shinn, *C.* (*Ceroliopoeum*) *laeta* (Vachal), *C.* (*Liopoeum*) *hirsutula* (Spinola), *C.* (*Hypomacrotera*) *callops* (Cockerell and Porter), *C. boharti* (Rozen), *C.* (*Nomadopsis*)

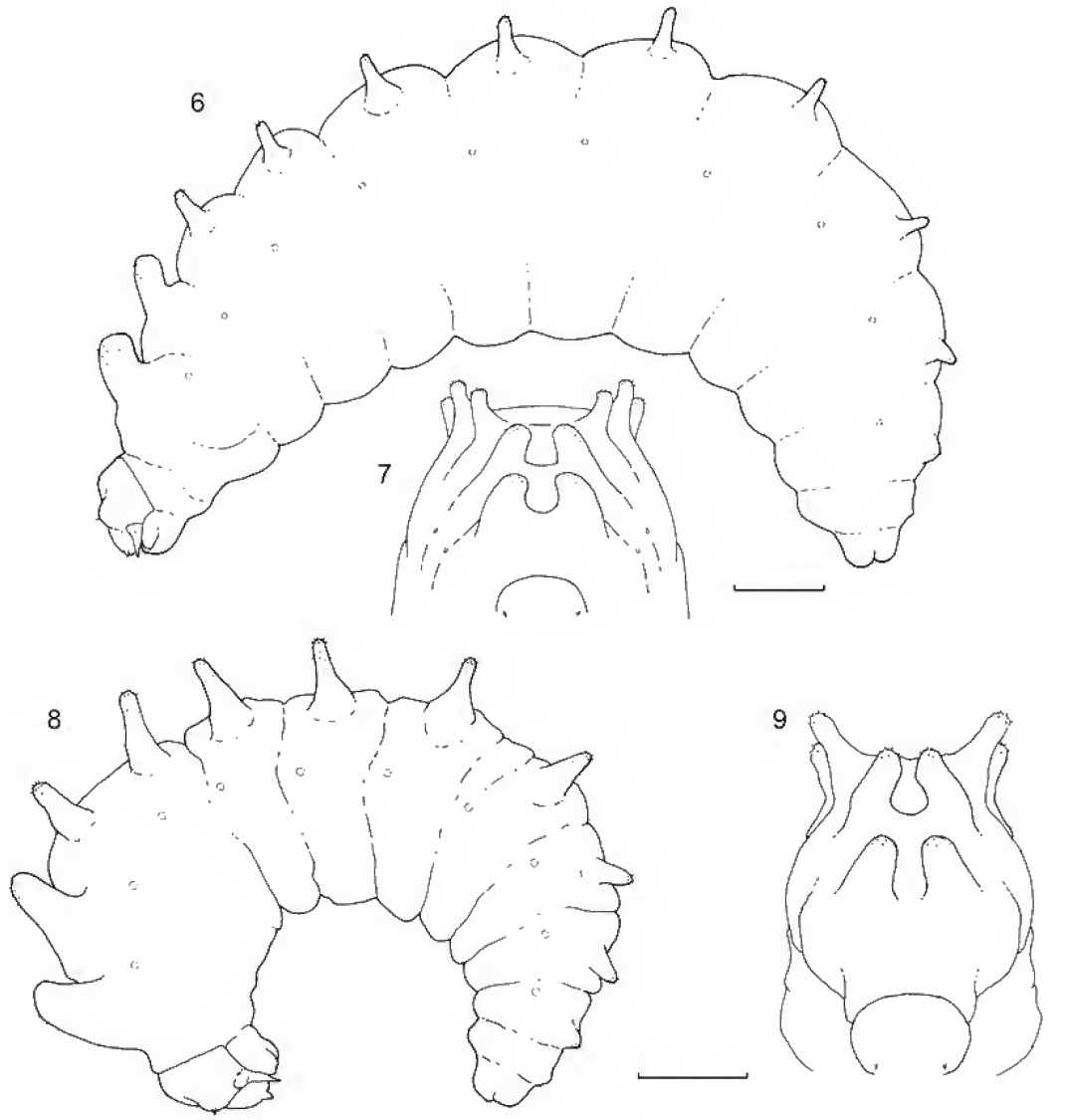


FIGURES 1–5. Camera lucida diagrams of cleared head capsules, lateral views, comparing antennal papillae with labral tubercles. **1.** *Spinoliella herbsti.* **2.** *Callonychium* (*C.*) *flaviventre.* **3.** *Callonychium* (*C.*) *petuniae.* **4.** *Callonychium* (*Paranychium*) *minutum.* **5.** *Arhysosage flava.* Scale bars = 0.5 mm.

linsleyi (Rozen), *C.* (*Macronomadopsis*) *zebrata bobbae* (Rozen), and *C.* (*Micronomadopsis*) *helianthi* (Swenk and Cockerell).

Diagnostic Comparisons of the *Spinoliella* Clade with Other Calliopsini and within the *Spinoliella* Clade

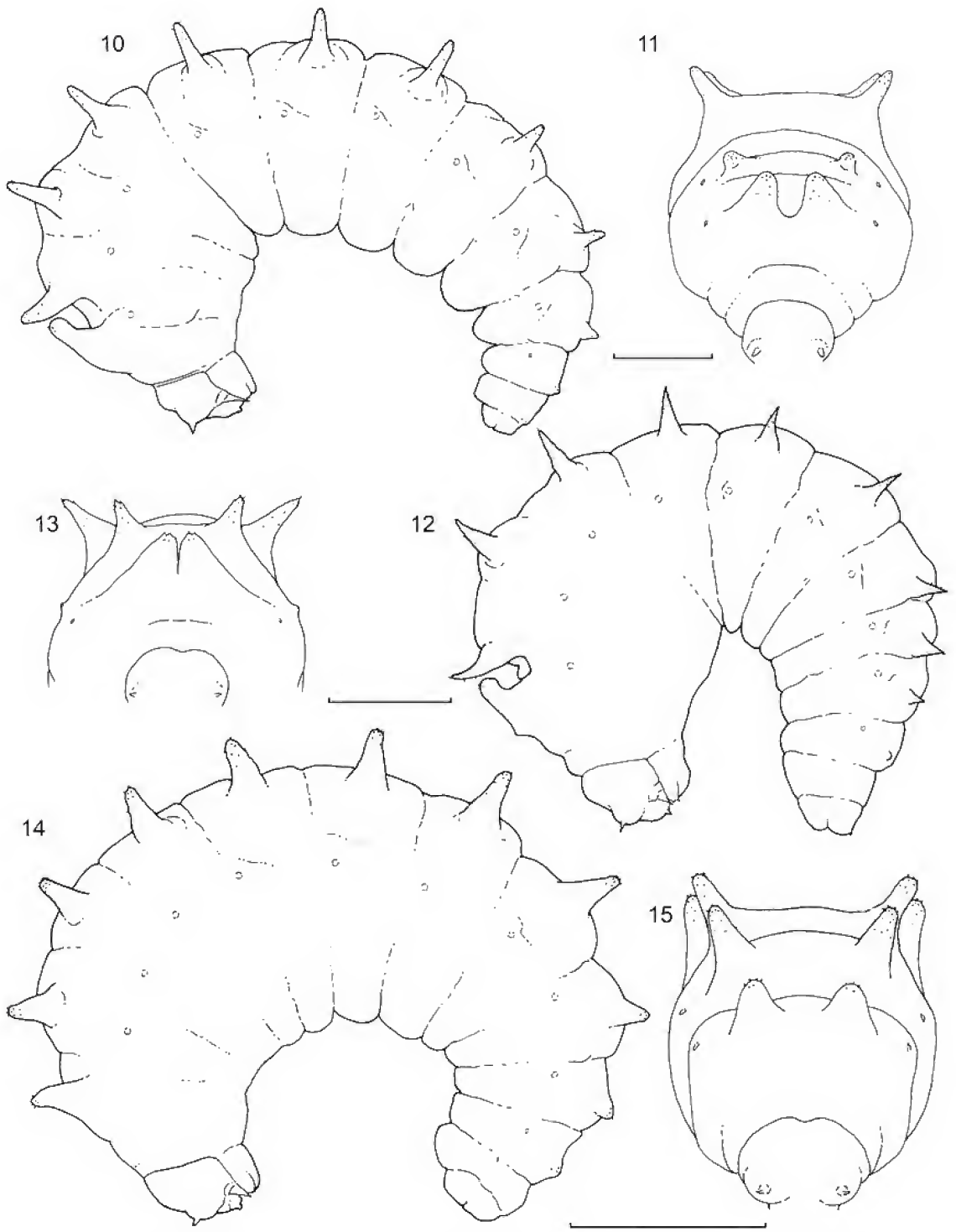
Mature larvae of *Acamptopoeum* and all known subgenera of *Calliopsis* have midbody paired dorsal tubercles that are short, with summits strongly transverse (i.e., transverse length



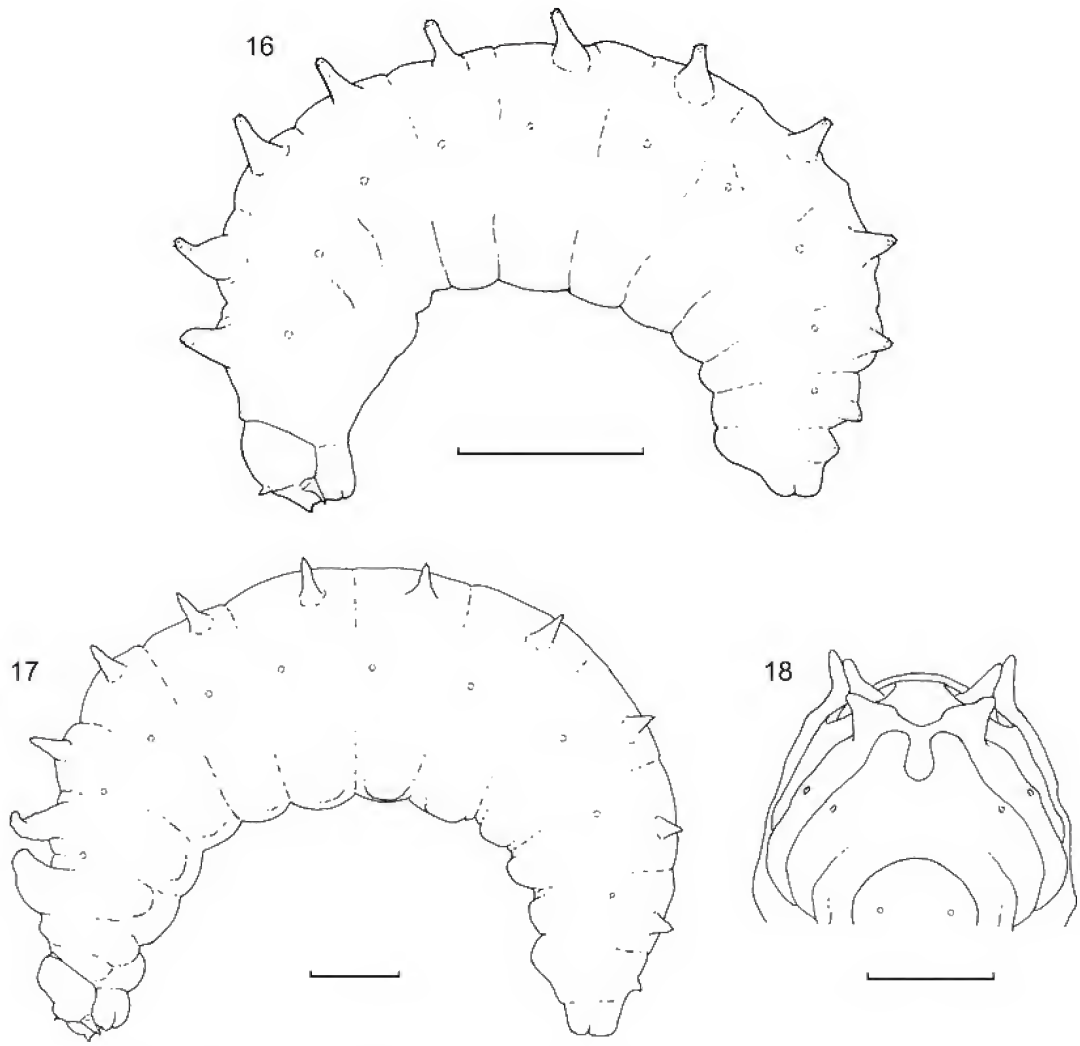
FIGURES 6–9. Camera lucida diagrams of mature larvae, lateral and frontal view, respectively; horizontal bars = 1.0 mm. **6, 7.** *Spinoliella herbsti.* **8, 9.** *Spinoliella maculata.*

much greater that anterior-posterior length, as seen from above) and quite similar to the paired dorsal tubercles of the pro- and mesothorax. By comparison, mature larvae of the species of *Spinoliella*, *Arhysosage*, and both subgenera of *Callonychium* treated here have midbody paired dorsal tubercles that are elongate, tapering to summits that are approximately circular in cross-section. These tubercles contrast particularly with those of the prothorax and to a lesser extent with those of the mesothorax, which tend to be larger and configured differently from the paired dorsal tubercles of the following segments (figs. 6–18).

Larvae of both species of *Spinoliella* have midbody tubercles somewhat blunter apically than those of *Callonychium* and *Arhysosage*. The antennal papillae of both species of *Spinoliella* are

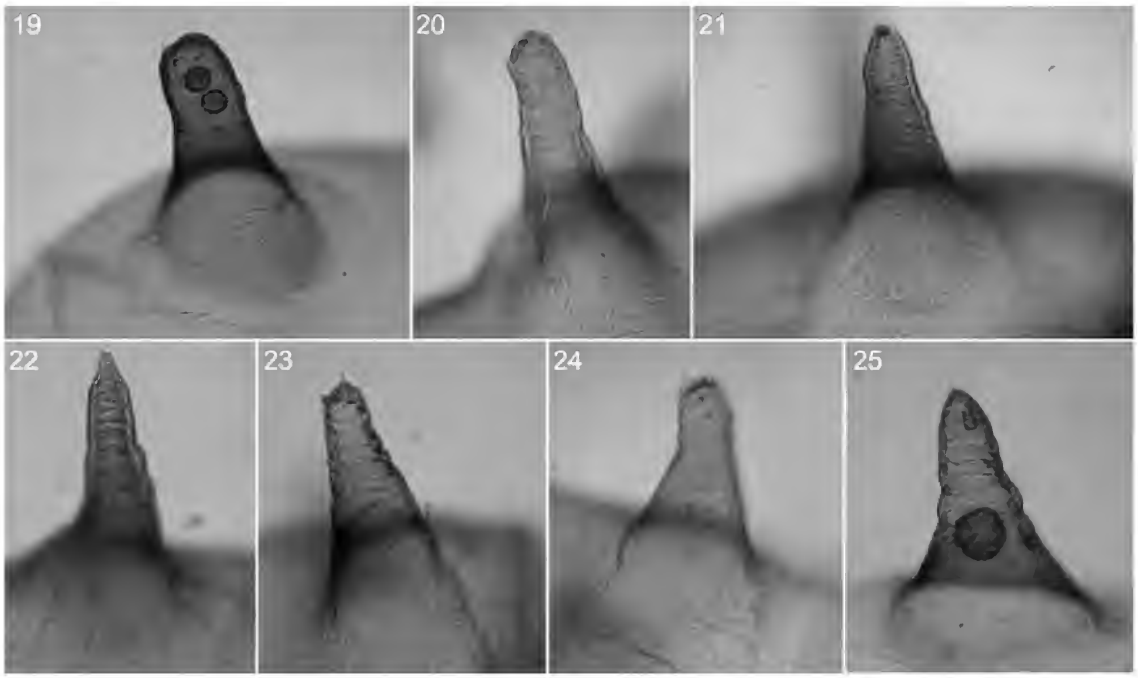


FIGURES 10–15. Camera lucida diagrams of mature larvae, lateral and frontal views, respectively; horizontal bars = 1.0 mm. **10, 11.** *Callonychium (C.) flaviventre.***12, 13.** *Callonychium* (*C.*) *petuniae.* **14, 15.** *Callonychium* (*Paranychium*) *minutum.*



FIGURES 16–18. Camera lucida diagrams of mature larvae; horizontal bars = 1.0 mm. **16.** *Callonychium* (*Paranychium*), n. sp., lateral view. **17, 18.** *Arhysosage flava*, lateral and frontal views.

moderately large, longer than their basal diameters, though clearly smaller than the labral tubercles. Thus they contrast with the antennal papillae of *Arhysosage flava*, which are quite short, their lengths clearly shorter than their basal diameters. Furthermore, the mesothoracic tubercles of both species of *Spinoliella* (as well as those of *Callonychium*) lean backward (figs. 6–16) and do not extend laterally nor apically twist backward as do those of *A. flava* (figs. 17, 18). This character of *A. flava* is unique among the three genera treated here, as is its serrated ventral apical mandibular edge and its small, short antennal papilla (resembling that found among species of *Calliopsis* and *Acamptopoeum*). While the antennal papillae of *Spinoliella* (fig. 1) are elongate compared with their basal diameters, the papillae are not as massive as those of any known *Callonychium*, all of which have papillae that are larger than the labral tubercles on the same specimen (figs. 2–4). This character then becomes a unique identifier of *Callonychium* compared with the other taxa. The reader



FIGURES 19–25. Photographs of cleared tubercle of abdominal segment 1 of each species, approximate side view. **19.** *Spinoliella herbsti.* **20.** *Spinoliella maculata.* **21.** *Callonychium* (*C.*) *flaviventre.* **22.** *Callonychium* (*C.*) *petuniae.* **23.** *Callonychium* (*Paranychium*) *minutum.* **24.** *Callonychium* (*Paranychium*), n. sp. **25.** *Arhysosage flava.*

should remember that these comparisons are among only a few representatives each of *Spinoliella, Arhysosage*, and *Callonychium* and may need reevaluation when more taxa are found.

Larval characters that define the clade are presented in **boldface** in the following description of the *Spinoliella* clade.

Mature Larvae of the *Spinoliella* Clade

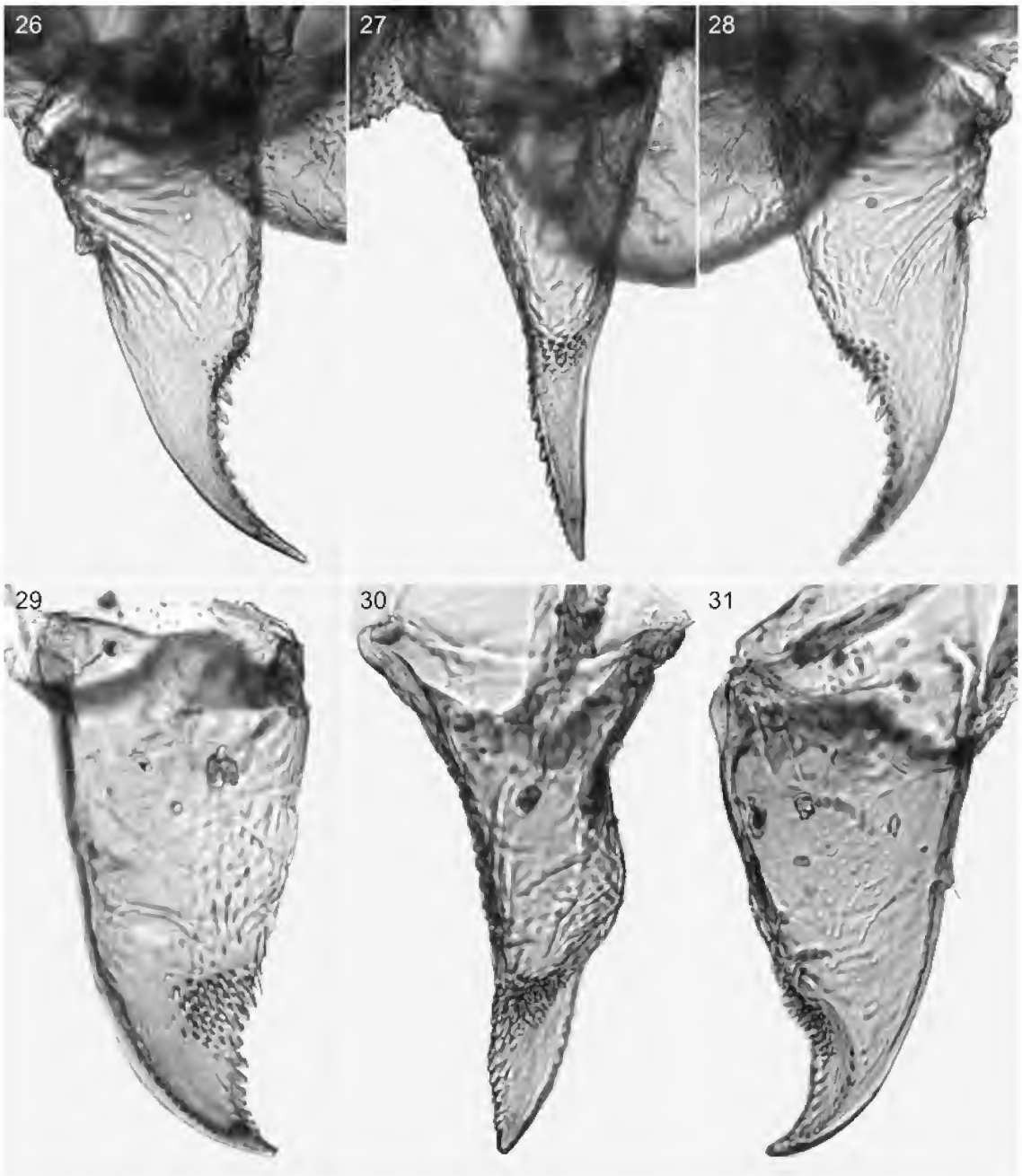
Description: **Head:** Integument with scattered sensilla some of which are setiform; dorsal surfaces of maxilla and hypopharynx densely spiculate; lateral surfaces of epipharynx spiculate; integument, particularly of maxillary apices, labial apex, and hypopharynx but also most of head capsule wrinkled. Integument unpigmented except antennal papilla, internal head ridges, and mandibular apices more or less pigmented.

Head size (figs. 8, 10, 12, 14, 16, 17) of postdefecating larva moderate compared with body size. Tentorium complete, including dorsal arms, but not robust. Anterior tentorial pit close to anterior mandibular articulation in frontal view; postoccipital ridge well developed; hypostomal ridge well developed; dorsal ramus of hypostomal ridge obscure; pleurostomal ridge moderately developed at both ends but weak in middle; epistomal ridge laterad of (below) anterior tentorial pit short; ridge between pits absent except faintly evident in *C. petuniae*; median longitudinal thickening of head capsule (coronal ridge) absent. Parietal bands faintly evident as integumental scars. **Antennal prominence strongly developed (figs. 1–5);** antennal disc as

seen on cleared specimen moderately small, its diameter somewhat less than one-half distance from its outer rim to center of anterior tentorial pit; **antennal papilla varying greatly in size, pigmented, tapering toward narrow apex, its height distinctly more than its basal diameter (figs. 1–4) except in *A. flava* (fig. 5),** bearing 3 to 4 sensilla. Vertex evenly rounded, without projections. Labrum projecting anteriorly about as far as clypeus in lateral view (figs. 1–5), with pair of distinct sensilla-bearing tubercles (figs. 1–5), which vary in size according to taxon.

Mandible elongate, in outer or inner views (figs. 27, 30) tapering evenly to narrowly pointed apex; dorsal apical edge with row of evenly spaced teeth that gradually increase in size toward cusp; ventral apical edge smooth, without teeth (fig. 27) except in *Arhysosage flava* (fig. 30), which has a finely serrate ventral apical mandibular edge; cuspal area scarcely produced, with cluster of teeth that are smaller than those of dorsal apical edge. Labiomaxillary region weakly projecting in lateral view (figs. 1–5), so that its apex exceeded by apex of labrum in lateral view; apex of maxilla projecting about as far as apex of labium in lateral view (figs. 1–5). Cardo, stipes not evident as sclerites; articulating arm of stipital sclerite not evident, although hypopharyngeal groove evident because of bulging surface above (hypopharynx) and below (labial apex); maxillary palpus (figs. 1–4) positioned at apex of maxilla, small, shorter than basal diameter, except in *A. flava* (fig. 5) in which palpus about as long as basal diameter. Labial pre- and postmentum indistinguishable (premental sclerite not evident); labial palpus small, scarcely projecting. Salivary opening small transverse slit curving upward at both ends, without lips. Hypopharynx protuberant, projecting forward, somewhat farther than labial apex in lateral view (fig. 1), except in *Callonychium* s.l., where it is recessed from labial apex.

Body: (figs. 6–25): Integumental vestiture of spicules and fine setae variable, but setae always short; **apices of all paired body tubercles with integument becoming faintly more sclerotized apically and having small seta-bearing swellings; these setae fine, tending to be erect.** Body form of predefecating larva moderately robust; **posterior abdominal segments tending to taper toward narrow abdominal apex (more obviously noticed on postdefecating forms (figs. 8, 10, 12, 14, 16), in which diminishing posterior part of abdomen contrasts sharply with robust anterior part of body;** intersegmental lines moderately weakly incised; dorsal intrasegmental lines not evident; paired dorsal tubercles present on all body segments except abdominal segment 10; **paired dorsal tubercles of prothorax and mesothorax large, but shape as well as degree and direction of slope variable (see Diagnosis); metathoracic paired dorsal tubercles tending to be similar to paired dorsal tubercles of abdominal segments 1–5, which are erect, apically slender, with apices circular in cross section; all paired dorsal body tubercles tending to have more or less conspicuous, scattered seta-bearing swellings on apical surfaces,** abdominal segment 9 not produced ventrally; abdominal segment 10 positioned medially on 9 as seen in lateral view (figs. 6, 8, 10, 12, 14,16, 17); anus apical on 10. Spiracles (figs. 32, 33) moderate in size to small, most on same specimens subequal in size except last pair tending to be smaller; on most species, spiracles on postdefecating forms tend to have narrow surrounding sclerite (indistinct in *S. maculata*), but those on predefecating larvae (*Spinoliella herbsti, Arhysosage flava*) without sclerites; peritreme (fig. 33) present, broad in radial width, which is somewhat larger than diameter of atrial opening though hard to evaluate without SEM micrograph; atrium projecting little beyond body wall, with



FIGURES 26–31. Microphotographs of cleared mature larvae. **26–28.** *Spinoliella herbsti*, right mandible, dorsal, inner, and ventral views. respectively, showing smooth ventral apical edge. **29–31.** *Arhysosage flava*, right mandible, dorsal inner, and ventral views, respectively, showing row of fine teeth along ventral apical edge.

low but distinct rim, globose; atrial wall smooth (without denticles or ridges); primary tracheal opening with collar; atrium in lateral view (fig. 32) small relative to width of subatrium; subatrium long, consisting of about 12–15 chambers. Sex recognition characters unknown.

Predefecating Larva and Biology of *Spinoliella herbsti* (Friese)

Figures 1, 6, 7, 19, 26–28, 32, 33

Diagnosis: The large, backward-tilting mesothoracic paired dorsal tubercles (figs. 6, 8) of the *Spinoliella herbsti* and *S. petuniae* separate them immediately from the other taxa in these three genera, as will the more slender but still elongate antennal papilla (fig. 1). Features have not been identified whereby larvae of the two species can be differentiated.

Material Studied: Six predefecating larvae: Chile: Coquimbo Region: Tilama, XI-02-1969 (J.G. Rozen).

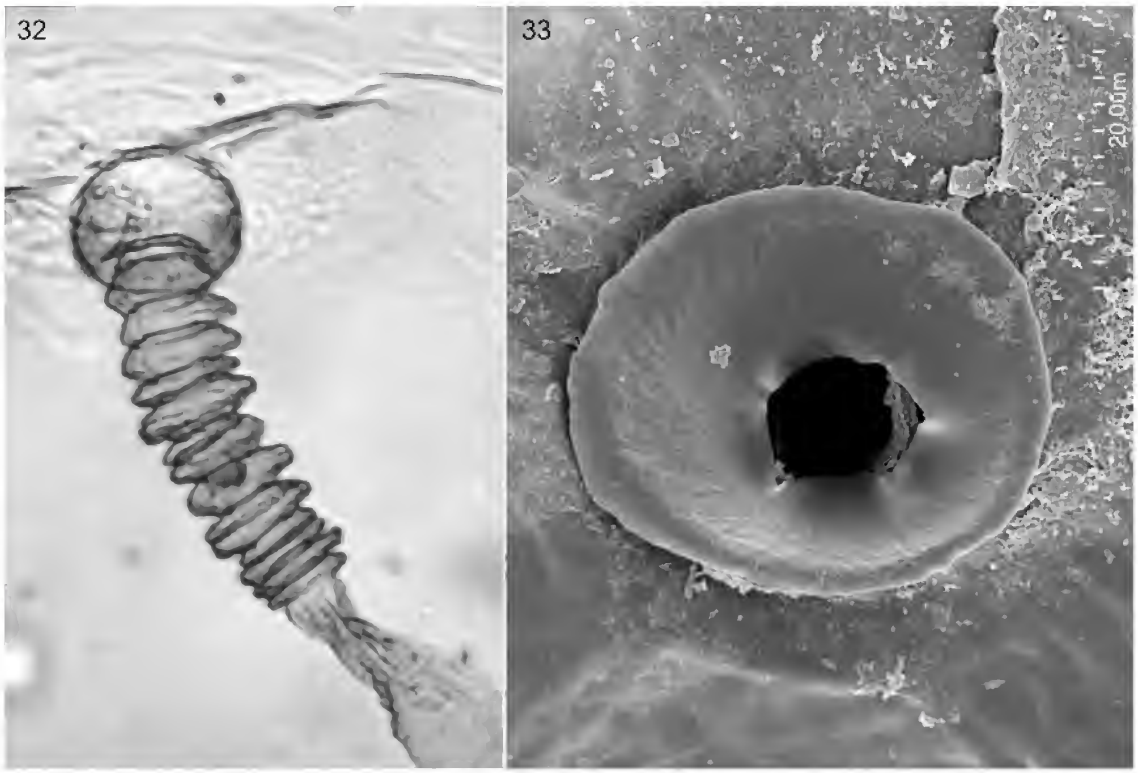
Remarks: Although the body form of the predefecating larva is quite robust, the more slender form of the postdefecating larva of *Spinoliella maculata*, described below (fig. 8), will likely be found also typical of postdefecating *S. herbsti*.

The internal head ridges and tentorium on one cleared specimen were considerably modified in that the tentorial bridge was bowed forward in association with the lower end of the postoccipital ridge, and the posterior part of the hypostomal ridge turned mesad and then bent forward. As a consequence, the lower posterior end of the parietal was also bent forward. Since this phenomenon occurred symmetrically on each side of the head capsule, this may be a normal change, perhaps an accommodation allowing the development of the pupal skeleton.

The function of the setae-bearing swellings on the paired dorsal tubercles (figs. 19, 20) is unclear. Initially I thought they might provide traction. Later, after discovering fine setae of similar length without associated swellings on the venter of abdominal segment 10, I considered that they might provide a sensory function. Observations on moving live larvae may provide answers.

Biological Notes: *Spinoliella herbsti*, *Calliopsis* (*Liopoeum*) *trifasciata* (Spinola), and *Liphanthus* (*L.*) *sabulosus* Reed nested in a barren area adjacent to an irrigated wheat field at Tilama, Coquimbo Region, Chile. The area, discovered on October 24, 1969, was excavated then and subsequently on November 2, 1969. Bordered by low trees on one side and by low vegetation elsewhere, the nesting area was unshaded during the main part of the day. Surface soil was dry, loose, and without stones but more compact just below the surface and moist from irrigation. Surface slope was about 10%, and pollen plants of none of the species grew adjacent to the site.

Nest Structure: A single burrow at the edge of a small raised area was identified when a female of *Spinoliella herbsti* entered it on October 24. Cells were located in the soil beneath in association with a meandering, obliquely descending, open burrow. Cells (table 1) were oriented horizontally and were narrowly pointed (i.e., not evenly rounded as in *Calliopsis*) at the posterior end. Their smooth surface gave no indication of a special lining. Closures were spirals, concave on the inside, and side tunnels were soil filled. Early provision loads were shaped into uncoated spheres; one incomplete load was 3.0 mm in diameter, while completed spheres were substantially larger (table 1). Homogeneously firm and moist on the inside, they were coated with a thick, shiny, clear, waterproof lining. Completed provisions rested near the center of the cell floor. Eggs were not found, but immobile young larvae, with heads pointed toward cell closures, rested on top of food spheres while eating provisions beneath their heads.



FIGURES 32, 33. Microphotograph of cleared spiracle of *Spinoliella herbsti*, side view, and SEM micrograph, showing broad peritreme, respectively.

A return to the site on November 2, 1969, yielded further information. Although it was impossible to follow any single nest, they consisted of at least four or five cells with the older cells nearer to the surface of the ground. Main burrows were soil filled most of the way down. When tested, cell walls readily absorb water droplets over the entire surface with the possible exception of the middle surface where food spheres rested. One larva was found lying on its dorsum while eating the remains of the provision on its venter, a clear indication that larvae reorient during the course of feeding.

Postdefecating Larva of *Spinoliella maculata* (Spinola)

Figures 8, 9, 20

Diagnosis: The single available larval specimen of *Spinoliella maculata* was partly destroyed after illustration (figs. 8, 9), so that details of its integument cannot be compared with those of *S. herbsti*. However, larval body forms of the two (figs. 6–9) were nearly identical except *S. maculata* was postdefecating and *S. herbsti* predefecating. Size difference between them was real (note: figs. 6–9 not to same scale), and the seta-bearing swelling on paired dorsal body tubercle were substantially more pronounced on *S. maculata* (fig. 20). All features in boldface in the description of larval *S. herbsti* are also characteristic of *S. maculata*.

Material Studied: One postdefecating larva: Chile: Valparaiso Region: Reñaca, X-22-94 (J.G. Rozen) collected as predefecating larva; preserved XI-9-94 as postdefecating larva.

Biological Notes: *Spinoliella maculata* was discovered nesting in an open sandy area at Reñaca, north of Valparaiso, Chile, on October 22, 1994, where a number of nests were examined. Nests (see table 1 for nest statistics) consisted of a short main tunnel that obliquely entered the nearly horizontal, unshaded ground and then branched into one or more long downcurving laterals each of which ended by abruptly turning and connecting with a single horizontal cell. Laterals were soil filled after cell closure. The entrance hole of the main burrow was crescentic in shape, presumably because of the loose, dry nature of the surface sand, which became moist below 5 cm. When excavating nests, females kicked sand from entrances with their hind legs, the tarsi of which exhibit a row of long setae on each side, thus forming a troughlike structure. At the posterior end of a cell, a 3.5 mm long, narrowly round extension (figs. 34, 35) providing the cell with a narrowly rounded outline similar to that of figure 47 but even more exaggerated. There was no visible special cell lining. All provision masses were spherical and thickly coated with a shiny, transparent, waterproof substance except for a smaller sphere, 2.6 mm in diameter, an indication that preliminary loads of provisions are shaped as well as the final food mass. An elongate, distinctly curved, smooth, whitish egg was attached by both ends to the top of the food sphere in the sagittal plane of the cell. It had a rounded front end and an elongate, pointed posterior end. The pollen source for this species was said to be *Cristaria* (Malvaceae) although Claude-Joseph (1926) had claimed it to be *Medicago sativa* L. (Fabaceae) and *Leucanthemum* (Asteraceae).

Mature Larvae and Biology of *Callonychium* (*Callonychium*) *flaviventre* (Friese)

Figures 2, 12, 13, 21

Diagnosis: Although mature larvae of *Callonychium* (*C.*) *flaviventre*, *C.* (*C.*) *petuniae*, *C.* (*Paranychium*) *minutum*, and *C.* (*P.*) n. sp. agree in many ways with those of *Spinoliella* and *Arhysosage flava*, they can immediately be recognized because their very large antennal papillae (figs. 2–4) are longer than the respective labral tubercles on the same specimen. All three genera have large antennal prominences. The more anterior paired abdominal tubercles of larval *Callonychium* (figs. 21–23) are apically slightly more tapering than those of *Spinoliella* (figs. 19, 20), which appear blunter owing in part to the prominent seta-bearing swellings.

The differences among the species of known larval *Callonychium* s.l., are subtle, as follows: *Callonychium* s.s.: antennal papilla directed nearly straight forward in lateral view (figs. 2, 3); body size larger; and prothoracic paired dorsal tubercles strongly directed backward (figs. 10, 12). *Callonychium* (*C.*) *petuniae* (figs. 12, 13) has more acutely pointed midbody paired dorsal tubercles whereas these tubercles of *C.* (*C.*) *flaviventre* (fig. 10) are more apically rounded. *Paranychium* (including *C. minutum*, *C.* n. sp., and an unknown species from Peru[2]) with

[2] Peru: La Libertad Dept.: La Quinta, 9 km. NNE Paiján, V-23-1996 (J.G. Rozen, A. Ugarte).

antennal papilla curving upward in lateral view (fig. 4); body size smaller; prothoracic paired dorsal tubercles nearly erect (figs. 14, 16), and mesothoracic paired dorsal tubercles little modified (figs. 14, 16).

Material Studied: Two postdefecating larvae: Argentina: Tucumán Province: 8 km SW Ticucho, collected as predefecating larvae III-25-1990; preserved IV-12-1990 (J.G. Rozen, A. Roig-Alsina). Five postdefecating larvae: Argentina: Tucumán Province: Amaichá del Valle, III-5, 11-1990 (J.G. Rozen, A. Roig-Alsina).

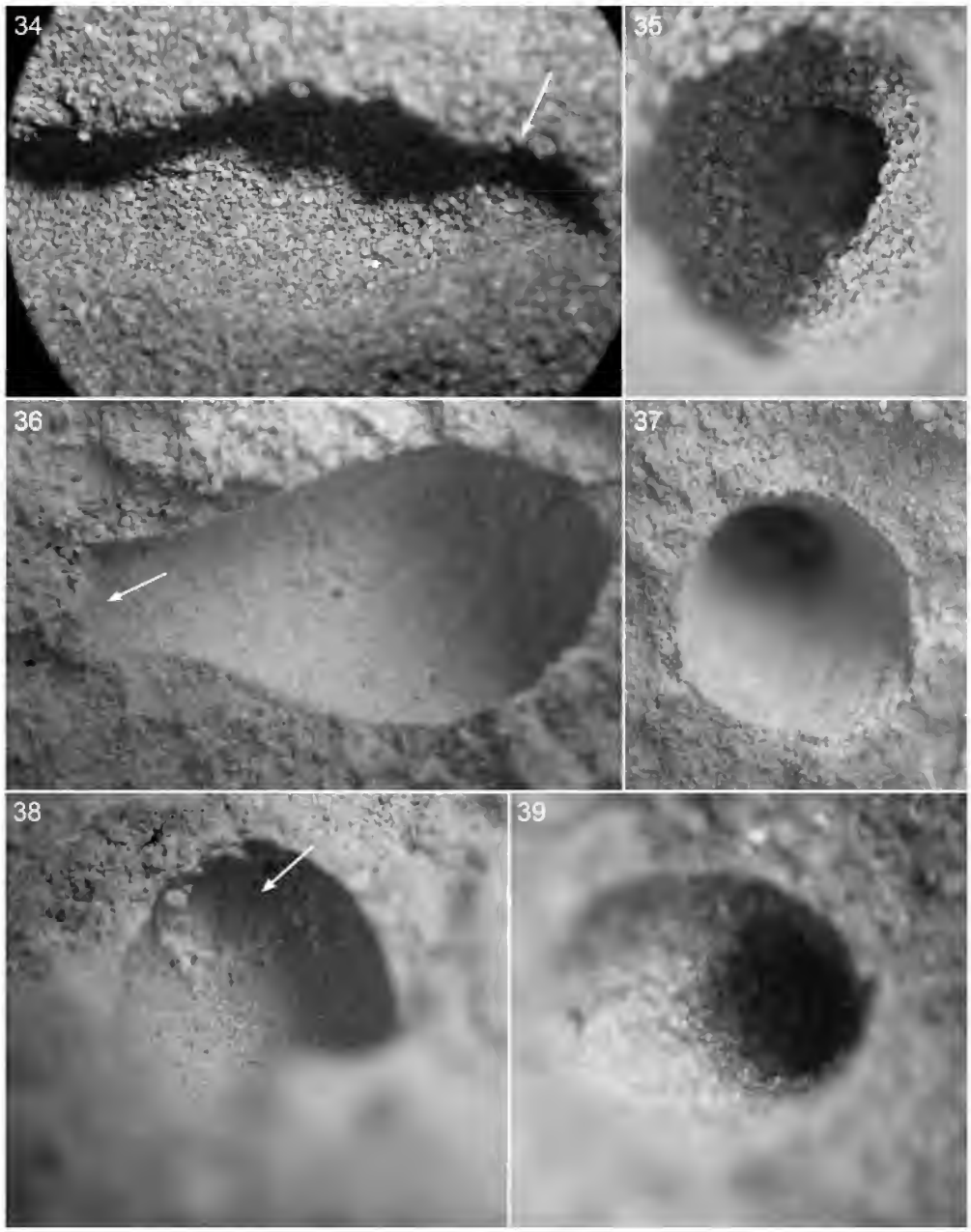
Biological Notes: The nesting biology of *Callonychium flaviventre* was investigated in Tucumán Province, Argentina, first at Amaichá del Valle a number of times between March 5 and 23 and then at 8 km southwest of Ticucho on March 25, all in 1990. At Amaichá del Valle the ground was dry and mostly horizontal on the surface with some large and small surface stones and moderately soft sand with stones below the surface, becoming more compacted below 3 cm. *Grindelia* (Asteraceae), the larval food source, grew 3–4 m away. However, the unusual nesting biology of the subgenus *Callonychium* was only fully understood after the nest of *C. flaviventre* was examined at the Ticucho site. There, because of the even soil texture, the shape of the cell and its entrance were fully explained, as described in the following paragraph. Cells were approximately 8 cm deep at the lower end of main tunnels that first penetrated the surface at a low angle and then descended with little twisting to the cell level.

Burrows at both sites descended in a meandering fashion and were often partly soil filled above but open farther down. All cells were essentially horizontal with an evenly curved posterior end and a front end that was consistently closed with a deeply recessed spiral closure providing a recessed, thimble-shaped space at the front of the cell (figs. 36, 37). It is in this recess (with a maximum diameter of a cell entrance and a lateral depth of 3–5 mm in several cases) where the defecating larva deposits its feces (fig. 38) and not at the cell's posterior end as in all other known calliopsines.

Cell dimensions in table 1 are presented separately for the Amaichá del Valle and Ticucho sites; cell lengths were measured from the posterior end of the cell to its front end but not including the open space created by the deeply concave closure in the entrance tunnel. Dimensions of food spheres for each site are also given; one presumably earlier supply was shaped as an uncoated sphere 2.8 mm in diameter. All completed provisions were approximately spherical and thickly coated with a clear, shiny, waterproof material.

The number of cells associated with a nest is uncertain. Only one was clearly found associated with a single burrow, but there is a possibility that a partly open burrow is not a main burrow but a long lateral and that each long lateral is filled after cell closure, and so goes undetected. This hypothesis was suggested by several nests of *Spinoliella maculata* found later in sandy soil near Valparaiso, Chile. This confusion can be resolved by monitoring a single nest under construction over a number of days.

Two slender, elongate, parallel-sided eggs (see table 1 for statistics) were observed at Amaichá del Valle. Each was attached by its posterior end to the top of the food sphere in the sagittal plane of the cell while the front end, pointing toward the cell front, was well



FIGURES 34, 35. Microphotographs of cells of *Spinoliella maculata*. **34.** Entire cell without closure, lateral view, with anterior end at left, showing elongate extensions (arrow) at posterior end. **35.** Posterior end of cell, inside view, showing entrance to posterior extensions. FIGURES 36–38. Microphotographs of cells of *Callonychium* (*C.*) *flaviventre*. **36.** Entire cell, lateral view, with anterior end at left, showing recessed cell closure (arrow) and space behind it where larval feces would be deposited. **37.** Front end of cell, inside view, showing closure end before defecations. **38.** Front end of another cell, inside view, after defecation (arrow). FIGURE 39. Microphotograph of front end of cell of *Callonychium* (*C.*) *petuniae*, inside view, showing deeply recessed spiral closure.

above the food surface. They were curved, whitish, and possessed a clear, shiny chorion. Possibly single attachments of panurgine eggs to provisions are seen with freshly deposited eggs. Later, as the embryo matures, the front end of the egg descends to contact the surface of the provisions, as has been suggested by other panurgines where both fresh and later ovipositions have been detected. This needs confirmation through sequential observations on a single egg during development.

Also at the Amaichá del Valle site several older immatures were encountered including a large larva on its dorsum with its front end close to the cell entrance while it fed on provisions, a fully fed predefecating larva also on it dorsum with its posterior end directed toward the cell entrance, and a pupa again on its dorsum and its metasoma directed toward the feces-filled entrance chamber.[3] Finally at the Ticucho site, two postdefecating larvae were found on their dorsa with their front ends directed toward the rear of the cell, their posterior ends pointed to the front, and the two entrance chambers were filled with feces. This series of observations confirmed that this species, like *Callonychium petuniae*, below, but unlike any other known panurgine, deposits its fecal material at the front of the brood chamber rather than at the rear.

Another remarkable feature of this species was the presence of a clear yellowish to amber-colored liquid detected between the paired prothoracic dorsal tubercles and the mesothoracic tubercles after larvae were finished feeding but not before. The source of this liquid and the role (if any) that it plays in the biology of this bee is unknown, but it has also been detected in the mature larva of *Callonychium* (*Paranychium*) *minutum* from the same locality: see Biology of *Callonychium* (*Paranychium*) *minutum*, below.

Larva and Biology of *Callonychium* (*Callonychium*) *petuniae* Cure and Wittmann

Figures 3, 12, 13, 22

This species was first studied at Vila Velha, Brazil, in 1971 and again in 2002. Gabriel A.R. Melo (e-mail: XII-14-12) stated that it is the only known species of *Callonychium* at this locality.

Diagnosis: See Diagnosis of *Callonychium flaviventre* and of *Spinoliella herbsti*.

Material Studied: Ten mature larvae: Brazil: Paraná: Vila Velha, XI-5-71 and XI-21-2002 (J.G. Rozen).

Biological Notes: The nesting biology of *Callonychium petuniae* was studied on two widely separated occasions at Vila Velha, Paraná, Brazil., the first on November 5, 1971, and again on November 23, 2002, although most of the following information came from the initial study. At that time the nesting site was along the side of one of the numerous sandy walkways that lead through the state park. The soil was composed of moderately coarse,

[3] Also at the Amaichá del Valle site the very first postdefecating larva recorded in the notes (III-05-1990) is depicted facing the feces-filled entrance of the cell. This presumably is a lapsus in recording since subsequent observation did not confirm it.

even-grained sand with no rocks and only a few small roots; dry on the surface, it was moist below 3 cm. The site was exposed to the sun during most of the day, was well elevated, contained a few scattered plants, and had a gently sloping surface. About 20–25 nests occurred within an area of 2 × 1 m. The same site was used for nesting by a species of *Lasioglossum* (*Dialictus*) (Halictidae). Neither in 1971 nor in 2002 was the pollen source identified at the site, although the species is known to be oligolectic on *Calibrachoa* (Solanaceae), a genus closely related to *Petunia* and perhaps also on *Petunia* (e-mail: G.A.R. Melo, XII-12-2012).

Nest entrances, irregularly spaced, were marked by conspicuous tumuli, 1.5–2.0 cm in diameter and as much as 1 cm in elevation. Most entrances were obscured by the tumuli but could be detected as eccentric depressions near the edges of tumuli. Pollen-laden females were occasionally seen returning from foraging, briefly landing and then observed "swimming" through tumuli to gain access to nest entrances. Occasionally the burrow entrance was exposed when the tumulus was on the downhill side. Among 10 nests excavated, all main burrows descended usually at about a 45° angle in a meandering fashion. Main tunnels (see table 1 for nest statistics) were filled at least at the surface but may have been only partly clogged with loose soil below. Nests consisted of a number of single cells connected to the main burrow by laterals 1 to 2 cm in length and 2.0 mm in diameter that were soil filled after cell completion. Cells had broadly rounded posterior ends like those of *Callonychium flaviventre* (fig. 36).

All cells were nearly horizontal and arranged singly. Provisions, coated with a conspicuous, clear, shiny material, were often only roughly spherical, as follows: 2.1 × 2.25 mm; 2.25 × 2.4 mm; 2.25 mm (spherical); and 2.6 mm (spherical), hence contrasting with the uniform spherical shapes of provisions of other calliopsine genera. Cell walls had a special lining somewhat darker than the surrounding sand but also had a coarse surface texture due to the coarseness of the sand. Cells were closed with earthen plugs, which in 1971 could not be observed because all four cell closures found were obscured by feces, but one was a deeply recessed spiral on a single preserved cell collected in 2002 (fig. 39). Early provision loads were shaped as a small, roughly spherical ball that was mealy moist as were the completed provisions on the inside. However, completed provision spheres were coated with a conspicuous, thick, shiny coating.

Two slightly curved eggs (table 1) were transparent white in color and had smooth shiny chorions. One was found attached by only its rear end to the provisions while its front end was elevated and directed toward the cell closure. In another cell a young larva sat on the top of the provisions and ate the provisions immediately below its head. One postdefecating larva rested on its dorsum with its anterior end toward the evenly round posterior end of the cell and with its posterior end directed toward the feces-filled cell closure.

Although observations were brief at Vila Velha, they suggested that females were active only the morning and very early in the afternoon. They were noted flying into the nesting site at 10 A.M., but by 1 P.M. few returning females were observed. The nesting site was not infested with parasitic bees, and furthermore bombyliids and triungulin larvae of meloids were not commonly observed in the area.

Postdefecating Larva and Biology of *Callonychium* (*Paranychium*) *minutum* Friese

Figures 4, 14, 15, 23

Diagnosis: See Diagnosis of *Callonychium flaviventre.*

Material Studied: Seven postdefecating larvae: Argentina: Tucumán Province: Amaichá del Valle, collected as predefecating larvae III-23-1990; preserved IV-12-90 (J.G. Rozen, A. Roig-Alsina).

Biological Notes: Observations on nests and nesting of *Callonychium* (*Paranychium*) *minutum* were undertaken at four localities in Argentina. The first two were in Catamarca Province, the first at Copacabana on November 28, 1989, and then at Belén on November 30, 1989. The following year, nests of this species as well as of *C.* (*C.*) *flaviventre* were discovered along the edge of a slightly sloping field at Amaichá del Valle, Tucumán Province, and studied on March 11, 20, and 23, 1990. The final site, 10 km west of Media Agua, San Juan Province, was studied October 30, 1991.

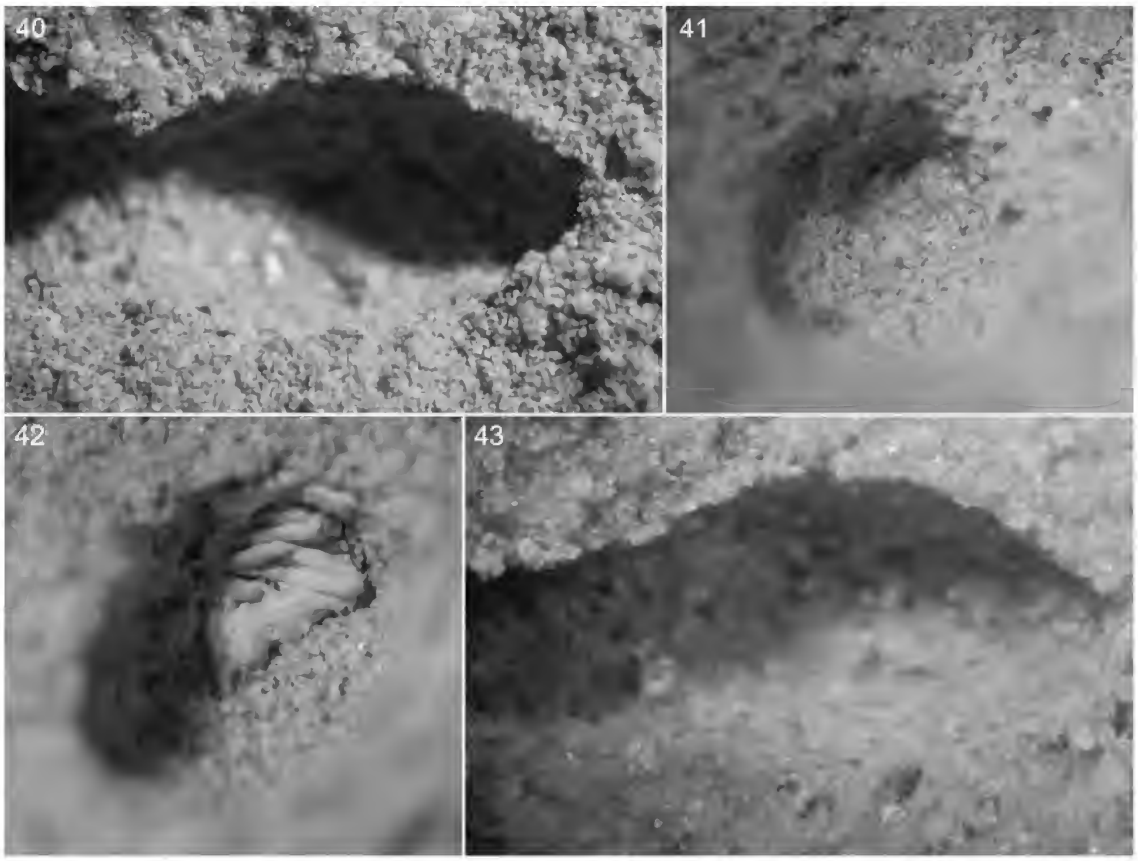
At all sites main tunnels were open, nearly vertical burrows usually with an abundant tumulus (see table 1 for nest statistics). Almost certainly each nest was constructed by a single female and possibly consisted of one cell per nest, although one nest seemed to have had two cells. Cells were oriented horizontally, without a visible special lining. Their shape was noteworthy in that they were elongate with an extended posterior end that, instead of being evenly curved as in most other panurgines, formed an acutely rounded terminus (figs. 40–42), which accommodated the fecal material voided by the mature larva. The inner surface of the cell closure was a rough, deeply concave spiral. Provisions (table 1) were spherical and coated with a clear, shiny surface; early loads were also spherical but substantially smaller and uncoated. Shiny, white, curved eggs were attached by their rear ends to the top surface of the provisions, while the anterior ends were raised above the provisions. A postdefecating larva found resting on its dorsum with head pointed toward cell closure and rear end directed toward the feces at the posterior end of the cell had clear yellowish to amber-colored liquid between the prothoracic and mesothoracic paired dorsal tubercles, as also noticed in the case of *Callonychium* (*C.*) *flaviventre.* The source and nature of this material is unknown. Males and females fly in copulo, often while females carry pollen on their hind legs.

The cleptoparasitic bee *Caenoprosopina holmbergi* Roig-Alsina (Apidae: Nomadinae: Caenoprosopidini) was collected in association with *Callonychium* (*Paranychium*) *minutum* at all of the sites mentioned above except for Media Agua, and its mature larvae were recovered and described from nests at Amaichá del Valle (Rozen and Roig-Alsina, 1991).

Mature Larva and Biology of *Callonychium* (*Paranychium*), Undescribed Species

Figures 16, 24

Luisa Ruz (e-mail: I-07-2013) after examining specimens reported that adults of this bee represent an undescribed species in the subgenus *Paranychium* close to *Callonychium* (*P.*) *minutum.*



FIGURES 40–42. Microphotographs of cells of *Callonychium* (*Paranychium*) *minutum*. **40.** Entire cell, lateral view with front end at left, showing narrowed posterior end of cell. **41.** Rear of cell, end view, before defecation. **42.** Rear of cell with feces. FIGURE 43. Microphotograph of cell of *Callonychium* (*Paranychium*), n. sp., lateral view, showing shape similar to that of *C. minutum* (fig. 40).

Diagnosis: The mature larva of this species cannot be easily distinguished from that of *Callonychium* (*Paranychium*) *minutum*, although the paired dorsal tubercles (fig. 24) are not quite as attenuated and evenly tapering as those (fig. 23) of *C. minutum*.

Material Studied: Ten mature larvae: Argentina: Jujuy Province: Tumbaya, XI-18, 19-1989 (J.G. Rozen, A. Roig-Alsina).

Biological Notes: This bee was discovered in copulo on blossoms of *Prosopis* (Fabaceae) and *Heliotropium* (Boraginaceae) and on the ground at Tumbaya, Jujuy Province, on November 18, 1989, and studied then and the following day. Nests were broadly scattered and those excavated on the first day were unusually deep and in a dry substrate (in table 1, top row refers to the dry substrate and bottom row, to moist substrate). In contrast those studied the next day 100 m away were in a very moist substrate and occurred at a depth of 4 cm, making nests of the species at this site the shallowest in this study. The dry area was removed from a source of water and the soil was sandy. The moist area was at the bottom of an artificial channel next to a river. In both situations, each nest was constructed by a single female, and the main tunnels were open and of about the same diameter. Cells were

arranged singly at the end of short laterals (ca. 1–3 cm long) that were soil filled after cell closure. Closures were deeply concave spirals on the inner surface. Burrows descended at about a 45° angle in the dry substrate, but the rate of descent seemed more variable under the moist condition. In both situations, cell orientation was the same (approximately horizontal); cell shape and size did not vary. Cells were narrowed at the posterior end (fig. 43), into which larval feces were deposited rather than on the posterior ceiling of the cell. In lateral outline cells seemed about as pointed at the front end as at the back end. Because copulating individuals were observed on both *Prosopis* and *Heliotropium*, possibly both plant species may be the larval food source. No cleptoparasites were associated with either of the nesting sites.

The most noteworthy discovery with this species was the substantial difference in cell depth that appears to be determined by the ground moisture level. Does this suggest cell depth is an adjustment for internal cell humidity, evidence that excavating moist substrate is more physically demanding, or...?

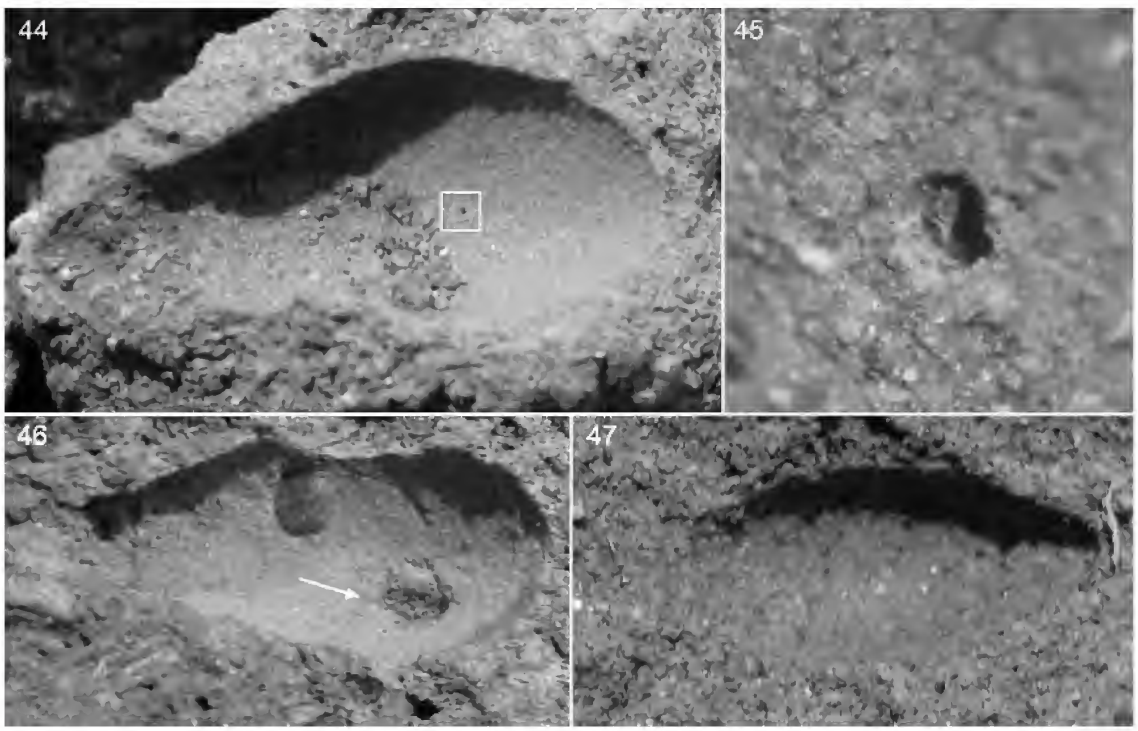
Predefecating Larva and Biology of *Arhysosage flava* Moure

Figures 17, 18, 25, 29–31

Diagnosis: The antennal papilla of the larva of *Arhysosage flava* is distinctly shorter than that of any other known larva in the *Spinoliella* clade and is slightly shorter than its own basal diameter. Furthermore, instead of having a smooth ventral apical edge to its mandible like others in the clade, its ventral apical mandibular edge is finely serrate. The paired dorsal mesothoracic tubercles are slender with the long axis of each directed more laterad (fig. 18) while its extreme apex curves posteriorly (fig. 17). The paired dorsal metathoracic and larger (more anterior) paired dorsal abdominal tubercles taper apically and have scattered short setae that arise from very small, inconspicuous swellings.

Material Studied: Two predefecating larvae: Argentina: Salta Province: Cruz Quemada, 40 km S. GRL. Güemes, XI-10-1989 (J.G. Rozen).

Biological Notes: The nesting site of *Arhysosage flava* and another, darker species, probably *A. ochracea* (Friese), was discovered at Cruz Quemada, 40 km south of General Güemes, Salta Province, Argentina, on November 10, 1989. The site of approximately 10 nests scattered over 2 m^2 of horizontal ground surface was first identified when an adult *Caenoprosopis* cleptoparasite was noticed sitting on a tumulus and matings of *A. flava* were observed in and around yellow flowers of the pollen plant, a low-growing cactus similar to or actually *Opuntia* (Cactaceae). Males flew rapidly from one cactus flower to another, alighted, and seemed to wait for females there. While in copulo, a male was seen to hold a female with his mandibles clasping her waist while she continued gathering pollen, a behavior also reported by Schlindwein and Wittmann (1995) for *A. cactorum* Moure. After being observed for a minute, they flew off in copulo, not unlike the mating behavior of females of *Calliopsis* (*Nomadopsis*) (Rozen 1958) except in the latter spiculate volsellae, not mandibles, seem involved with attachment. Nests appeared in early stages of construction; none yet had cells.



FIGURES 44–46. Microphotographs of cells of *Arhysosage flava*. **44.** Entire cell, lateral view, showing somewhat round rear. **45.** Close-up of insertion hole of egg of cleptoparasite *Caenoprosopis crabronina* identified by rectangle in figure 44. **46.** Cell in lateral view exhibiting rough pit (arrow) gouged in cell wall presumably by female *A. flava* removing egg of *Caenoprosopis crabronina*. FIGURE 47. Microphotographs of cell of *Arhysosage bifasciata*, lateral view, showing narrowly rounded rear.

When the site was again visited on November 20, 1989, four nests of *Arhysosage flava* (see table 1 for statistics) were excavated. All were moderately shallow with cells occurring singly (i.e., not in linear series) short distances away (ca. 1–2 mm) from meandering, open main burrows. Cells, horizontal to sloping backward 10°, had rather evenly curved, perhaps only slightly narrowed posterior ends (fig. 44), and cell walls were fine grained and nonwaterproof when tested. However, importantly, the cell floor possessed a water-retardant area 5 mm long and 4 mm wide on its lower surface; see Discussion. Provisions were formed into a sphere, on top of which eggs were placed and young larvae ate; predefecating larvae were found resting on their dorsa on the cell floor with their heads pointed toward cell closures. Provisions were thickly coated with a clear, waterproof material.

The cleptoparasitic bee *Caenoprosopis crabronina* Holmberg was found attacking nests of *Arhysosage flava* at this site (Rozen and Roig-Alsina, 1991). Figure 44 shows not only the shape of the rear of the cell, but also the egg insertion hole of the cleptoparasite, which embedded her egg in the cell wall, so that only the anterior end is seen (fig. 45) along with marks created by the host female where she excised a cleptoparasite egg (fig. 46) presumably with her mandibles.

Biology of *Arhysosage bifasciata* (Friese)

After the above observations of *Arhysosage flava* were made, a small group of nests of *A. bifasciata* (Friese) were excavated at El Desmonte, Catamarca Province, Argentina, on November 23, 1989, and on the following day (see table 1 for statistics). Several nest openings occurred under or at the edges of surface rocks; others were in the open. All nests (total of 6) were currently under construction and had diagonally descending, meandering, open main tunnels. No nest had more than two cells, but since no larvae were found, all nests were presumably undergoing development. Cells were horizontal or nearly so, and provisions were spherical and coated when complete. Two incomplete, roughly spherical provisions were much smaller (e.g., 2.9 mm in diameter) and uncoated. Perhaps the most significant difference between *A. flava* and *A. bifasciata* was a difference in cell shape. Instead of being somewhat rounded as in *A. flava* (fig. 44), the posterior end of the cell of *A. bifasciata* was more drawn out and therefore narrowly rounded (fig. 47). Since no larvae were yet present, it can not be determined whether larval feces are placed in this elongation, although this is a likely assumption.

DISCUSSION

The function of the setae-bearing swellings on the paired dorsal tubercles is unclear. Initially I thought they might provide traction (related to larval movement, see below), but after discovering fine setae of similar length, but without associated swellings on the venter of abdominal segment 10, I considered that they might have a sensory function. Observations on moving live larvae may suggest answers.

In the rest of this section I compare what is known about the biology and larval anatomy of the species of *Spinoliella*, *Callonychium*, and *Arhysosage* with one another and with life history attributes of other Calliopsini. Clearly some (and possibly all) species of *Spinoliella*, *Callonychium*, and *Arhysosage* fly while in copulo, as reported earlier for *Callonychium* (*C.*) *petuniae* (Cure and Wittmann, 1990), *C.* (*Paranychium*) *chilense* (Toro, 1985), *Arhysosage cactorum* (Schlindwein and Wittmann, 1995), a Brazilian species of *Arhysosage* soon to be described (Ramos, in press), and, as stated above, for *C.* (*P.*) *minutum*, *C.* (*P.*) n. sp., and *A. flava.* Although no mating observations were recorded for *Spinoliella*, *C.* (*C.*) *flaviventre*, or *A. bifasciata,* it would not be surprising if they were capable of such behavior considering what is known about related species. However, among all bees, few are known to fly in copulo. As one anonymous reviewer commented "the correlate of this behavior—extreme polyandry—seems to be common in the Panurginae, possibly universal in the Calliopsini, but rare elsewhere among bees."

All *Spinoliella*, *Callonychium*, and *Arhysosage* about which we have information are ground nesting (as are all known Andrenidae[4]), with single females working alone to construct shallow nests in which they excavate horizontal cells, and, like all other Andrenidae, none are known to spin cocoons. Nesting surfaces are generally level and more or less exposed to the sun. The cell wall of these taxa has little strength, appearing no more consolidated than the surrounding

[4] The one reported exception is *Perdita opuntiae* Cockerell, which was stated to nest in sandstone by Custer (1928), but see Bennett and Breed (1985).

soil. Another shared feature is the relatively smooth surface of the cell wall, which lack a visible lining, determined by microscopic examination, and which often allows water absorption when tested with water droplets. However, the discovery of a water-retardant area restricted to the lower part of the cell of *Arhysosage flava* where the food sphere rests suggests that more thorough testing should be done on all species.

The female of each species provisions the cell with pollen mixed presumably with nectar, forming the mass into a firm sphere after every foraging trip. After the final foraging trip, she coats the sphere with a transparent, reflective layer of material that is waterproof, as judged, for example, by spheres remaining intact even after years of being preserved in Kahle's solution. The source of the material is unknown, and its function is uncertain, though probably pertaining to humidity control in a cell lacking a waterproof lining.

Curved, white, shiny, elongate eggs (see table 1 for dimensions) are deposited on or near the top of the food spheres with all Calliopsini. Interestingly, eggs of species of *Callonychium* (egg deposition unknown for *Spinoliella* and *Arhysosage*) were attached by only their posterior ends with the front ends not touching the food. Sequential observations should be made to determine if or when a freshly deposited egg gradually bends downward prior to hatching so that its anterior end comes in contact with the provisions.

All early instars were positioned lengthwise on the provisions in the sagittal plane of the cells where their eggs had been deposited, as is characteristic of all known Panurginae (Michener, 2007). When approximately halfway through eating, larvae reorient so that they are lying with their dorsal surfaces on the cell floor and their anterior ends are directed toward the cell rear. There is no evidence that a larva crawls on the food mass at any time; its body always remains in the same position relative to the partly eaten food, even though the larva changes its direction in the cell and comes to rest on its dorsum while supporting the partly eaten food sphere on its venter.

Larvae have not been observed while they are repositioning. A likely hypothesis is that after eating part of the provisions and increasing in size, their paired dorsal body tubercles come in contact with the cell ceiling above. Presumably by some use of these tubercles against the cell surface (perhaps caused by repeatedly bending the forward part of their body downward to reach the receding food surface), they forward-somersault (presumably slowly) carrying the provisions with them. They thus come to rest facing the cell rear and consume the remaining food.[5] However likely this hypothesis, it is incomplete because it does not explain the use of the exaggerated prothoracic and mesothoracic paired dorsal tubercles of the *Spinoliella* clade, since larvae of *Calliopsis*, without exaggerated tubercles, also reposition themselves similarly and when doing so, carry uneaten provisions with them (e.g., Rozen, 1958).

Most of these features are characteristic of other Calliopsini, with the possible exception that coating of the provision may possibly be reduced or even absent among a few taxa, and occasionally cells in short linear series have been reported for panurgines (e.g., Rozen, 1958:

[5] Although the inability of young larval instars to crawl on their venters seems not only characteristic of Calliopsini and some other groups of Panurginae, it apparently is not true of all. *Panurginus potentillae* (Crawford) is thought to be able to crawl and exhibits anatomical features for doing so (Rozen, 1967).

fig. 65).[6] Of interest is the fact that similar coatings on provisions are also known for the genus *Perdita*, species of which also shape spherical food masses (Rozen, 1967; Danforth, 1989), but such coatings are apparently absent in *Macrotera* (Danforth, 1991; Neff and Danforth, 1992). Although this sharing suggests a synapomorphy, coated larval food spheres, though an uncommon phenomenon among most bees, have also been reported in the Rophitinae (Halictidae) (Eickwort et al., 1986; Rozen, 1993; Rozen and Özbek, 2008).

Some ethological features found in *Spinoliella*, *Callonychium*, and *Arhysosage*, however, are unusual, if not unique, and relate to placement of larval feces, cell shape, and configuration of the closure spiral. This is most clearly demonstrated by the two species in the subgenus *Callonychium* and the two in the subgenus *Paranychium*. Brood chambers of *C. flaviventre* (fig. 36) and *C. petuniae* have an evenly rounded posterior end in lateral view not unlike chambers of *Calliopsis* (Rozen, 1967: fig. 9; Shinn: 1967: fig. 155) and *Acamptopoeum* (Rozen and Yanega, 1999: figs. 1, 2). *Calliopsis* has a cell closure (Rozen, 1967: fig. 9) only slightly concave on the inner surface and places feces on the upper rear cell wall (as is characteristic of most other panurgines, Rozen, 1967: fig. 13; Shinn, 1967: fig. 155). However, both species of subgenus *Callonychium* have a recessed, deeply concave cell closure (figs. 36, 39) and in the empty space behind the closure at the front end of the cell, they deposit their larval feces (fig. 38). Three postdefecating larvae and one pupa of *C. flaviventre* and a postdefecating larva of *C.* (*C.*) *petuniae* were discovered on their dorsa with posterior ends directed toward cell closures.[7] Thus, evidence is strong that at least some emerging adults will need to reverse directions in their cells at time of egress.

The two species of *Paranychium, C.* (*P.*) *minutum* and *C.* (*P.*) n. sp., have the posterior end of the brood cell prolonged and narrowly rounded (figs. 40, 43) so as to appear semipointed, and the feces are discharged in this extra space at the rear of the cell (fig. 42), although their cells also have a recessed, deeply concave spiral closure, as do those of the subgenus *Callonychium*. Because postdefecating larvae rest on their dorsal surfaces with their anterior ends directed toward the closures, they must reorient in the brood cell after completing their food supply before they start defecating, a characteristic of *Calliopsis* as well.

Complete information on cell shape relative to feces placement is unknown for *Spinoliella* and *Arhysosage*, but cell shapes for *S. herbsti* and *S. maculata* (fig. 34) are available; both have unusual posterior extensions, although those of *S. maculata* may be even more exaggerated than those of *S. herbsti*. Likely that is where the feces will be found. For *Arhysosage*, evidence is less revealing. With *A. flava* the rear cell contour is perhaps faintly narrow on one specimen, but the front end of the cell shows a recessed closure. Until a cell bearing deposited feces is uncovered, the situation remains moot. In the case of *A. bifasciata* (fig. 47), the posterior end of the cell is elongate with a suggestion that the rearmost surface is less compacted, hinting that it might be more absorptive than the cell wall elsewhere.

[6] As pointed out by an anonymous reviewer: "The construction of cells in extended linear series by *Calliopsis persimilis* (Cockerell) seems to be a significant contributing factor to the very high daily cell completion rate of this species (Danforth, 1990)."

[7] A single postdefecating larva of *C. flaviventre* was recorded facing the opposite direction, suggesting either an error of observation or a variable behavior pattern.

This paper is a result of a series of short studies (nest excavations) usually lasting from an hour or two up to several days, gathered over numerous field trips to Chile, Argentina, and Brazil. These observations offer a degree of new information about the behavior of these bees, but also require further explanation. It seems plausible that the exaggerated paired dorsal body tubercles play a role enabling large larvae to reorient in their cells, and the tapering posterior of the larva may relate to placement of feces either in the narrow vestibule at the front of the cell (*Callonychium*) or the narrow posterior extension of the brood cell (*Paranychium*, *Spinoliella*). Certainly, additional fieldwork resulting in studies of more species will be useful in determining the extent to which these differences are taxonomically important. However, to explore adaptive functioning of the paired dorsal tubercles and fecal placement will probably require sequential observations on single developing live larvae in containers simulating the confines (shape) of brood chambers in a stable laboratory environment. Such studies may be difficult, but even one successful one will be invaluable.

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